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FILE 'HOME' ENTERED AT 20:27:06 ON 16 FEB 2003)

     FILE 'BIOSIS, CAPLUS, SCISEARCH, LIFESCI, EMBASE' ENTERED AT 20:27:54 ON
     16 FEB 2003
L1           153 S D-AMINOACYLASE
L2             0 S L1 (A) ZINC

     FILE 'CAPLUS' ENTERED AT 20:29:14 ON 16 FEB 2003
L3            65 S L1
L4             0 S L2
L5             0 S L1 (A) ZINC
L6             0 S L1 (A) ZINC ION
L7             4 S AMINOACYLASE (A) ZINC

     FILE 'BIOSIS, SCISEARCH, LIFESCI, EMBASE' ENTERED AT 20:37:24 ON 16 FEB
     2003
L8             0 S L7

     FILE 'BIOSIS' ENTERED AT 20:37:46 ON 16 FEB 2003
L9             0 S L7

     FILE 'SCISEARCH' ENTERED AT 20:38:03 ON 16 FEB 2003
L10            0 S L7

     FILE 'LIFESCI' ENTERED AT 20:38:21 ON 16 FEB 2003
L11            0 S L7

     FILE 'EMBASE' ENTERED AT 20:39:03 ON 16 FEB 2003
L12            0 S L7

     FILE 'USPATFULL, JAPIO, EUROPATFULL, PATOSWO' ENTERED AT 20:39:53 ON 16
     FEB 2003
L13            0 S L7
L14           44 S D-AMINOACYLASE
L15            0 S L14 (A) ZINC
L16            0 S USPAT

     FILE 'USPATFULL' ENTERED AT 20:41:37 ON 16 FEB 2003
L17           15 S L14
L18            0 S L17 (A) ZINC
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L17 ANSWER 2 OF 15 USPATFULL
AN 2003:33332 USPATFULL
TI D-aminoacylases, method for producing the same, and method for producing
D-amino acids using the same
IN Mitsuhashi, Kazuya, Ibaraki, JAPAN
Yamamoto, Hiroaki, Ibaraki, JAPAN
Matsuyama, Akinobu, Ibaraki, JAPAN
Tokuyama, Shinji, Shizuoka, JAPAN
PA Daicel Chemical Industries, Ltd., Osaka, JAPAN (non-U.S. corporation)
PI US 6514742 B1 20030204
AI US 1999-361901 19990727 (9)
PRAI JP 1998-228636 19980729
DT Utility
FS GRANTED
EXNAM Primary Examiner: Prouty, Rebecca E.
LREP Fish & Richardson P.C.
CLMN Number of Claims: 4
ECL Exemplary Claim: 1
DRWN 9 Drawing Figure(s); 5 Drawing Page(s)
LN.CNT 1048

L17 ANSWER 2 OF 15 USPATFULL

AB **D-aminoacylase** derived from fungi is provided. The fungi capable of producing **D-aminoacylase** include those belonging to the genus Hypomyces, Fusarium, Auricularia, Pythium, and Menisporopsis. The fungal **D-aminoacylase** is useful for efficiently producing D-amino acids from N-acetyl-D-amino acids.

7 ANSWER 3 OF 15 USPATFULL
AN 2002:272914 USPATFULL
TI **D-aminoacylase** and gene encoding the same
IN Mitsuhashi, Kazuya, Tsukuba-shi, JAPAN
Yamamoto, Hiroaki, Tsukuba-shi, JAPAN
Matsuyama, Akinobu, Tsukuba-shi, JAPAN
Tokuyama, Shinji, Shizuoka-shi, JAPAN
PI US 2002151035 A1 20021017
AI US 2001-770517 A1 20010126 (9)
PRAI JP 2000-19080 20000127
JP 2000-150578 20000522
DT Utility
FS APPLICATION
LREP JANIS K. FRASER, FISH & RICHARDSON P.C., 225 Franklin Street, Boston, MA, 02110-2804
CLMN Number of Claims: 13
ECL Exemplary Claim: 1
DRWN No Drawings
LN.CNT 1570

17 ANSWER 3 OF 15 USPATFULL

AB The present invention provides the **D-aminoacylase** -encoding gene derived from Hypomyces mycophilus, a filamentous fungus, the polypeptide encoded by the gene, and the homologues thereof. The **D-aminoacylase** of the present invention is capable of producing D-tryptophan from N-acetyl-D-tryptophan. D-tryptophan is useful as a medicinal raw material or the like.

17 ANSWER 8 OF 15 USPATFULL
AN 2000:24497 USPATFULL
TI **D-aminoacylase**
IN Tokuyama, Shinji, Shizuoka, Japan
PA Daicel Chemical Industries, Ltd., Osaka, Japan (non-U.S. corporation)
PI US 6030823 20000229
AI US 1999-268941 19990316 (9)
PRAI JP 1998-89246 19980317
JP 1999-35620 19990215
DT Utility
FS Granted
EXNAM Primary Examiner: Lankford, Jr., Leon B.
LREP Fish & Richardson P.C.
CLMN Number of Claims: 7
ECL Exemplary Claim: 1
DRWN 10 Drawing Figure(s); 6 Drawing Page(s)
LN.CNT 779

17 ANSWER 8 OF 15 USPATFULL

AB A novel **D-aminoacylase** was derived from a microorganism belonging to the genus Sebekia. This enzyme is useful for producing D-amino acids from N-acetyl-DL-amino acids on an industrial scale.

17 ANSWER 11 OF 15 USPATFULL
AN 1999:72467 USPATFULL
TI **D-aminoacylase**
IN Tokuyama, Shinji, Shizuoka, Japan
PA Daicel Chemical Industries, Ltd., Osaka, Japan (non-U.S. corporation)
PI US 5916774 19990629
AI US 1998-122386 19980724 (9)
PRAI JP 1997-206288 19970731
JP 1998-141932 19980522
DT Utility
FS Granted
EXNAM Primary Examiner: Wax, Robert A.; Assistant Examiner: Srivastava, Devesh
LREP Fish & Richardson P.C.
CLMN Number of Claims: 8
ECL Exemplary Claim: 1
DRWN 9 Drawing Figure(s); 8 Drawing Page(s)
LN.CNT 819

17 ANSWER 11 OF 15 USPATFULL

AB This invention provides a novel **D-aminoacylase** and a method for producing said enzyme, and also a method for producing D-amino acids using said aminoacylase. **D-aminoacylase** of the invention having novel properties can be derived from a microorganisms belonging to genus Amycolatopsis. The use of the enzyme enables industrial production of D-amino acids.

7 ANSWER 1 OF 4 CAPLUS COPYRIGHT 2003 ACS
AN 1995:54388 CAPLUS
DN 122:127410
TI Effect of zinc ion on conformation and its stability of aminoacylase
AU Zhang, Tong; Zhoo, Haimeng
CS Dep. Biol. Sci. Biotechnol., Tsinghua Univ., Beijing, 100084, Peop. Rep. China
SO Shengwu Wuli Xuebao (1994), 10(2), 198-202
CODEN: SWXUEN; ISSN: 1000-6737
DT Journal
LA Chinese
CC 7-5 (Enzymes)
AB The effect of Zn2+ on the secondary structure of aminoacylase was studied by CD and deconvolved FTIR spectroscopy. After removal of Zn2+, the contents of enzyme ordered secondary structure decreased. Fluorescence emission spectrum showed that the emission max. of the apoenzyme, as compared to the holoenzyme, was red-shifted from 335 to 336.5 nm, indicating the occurrence of some unfolding of the tertiary structure of the apoenzyme. The stability of the apoenzyme in detergent decreased markedly. Thus, Zn2+ helps to maintain the active site of aminoacylase in a specific, stable conformational state.
ST **zinc aminoacylase** conformation stability
IT Enzyme functional sites
(zinc ion contribution to the conformation and stability of aminoacylase)
IT Conformation and Conformers
(secondary, zinc ion contribution to the conformation and stability of aminoacylase)
IT 7440-66-6, Zinc, properties 9012-37-7, **Aminoacylase**
RL: PRP (Properties)
(**zinc** ion contribution to the conformation and stability of aminoacylase)

L7 ANSWER 2 OF 4 CAPLUS COPYRIGHT 2003 ACS
AN 1993:665227 CAPLUS
DN 119:265227
TI A comparison of zinc(II) and cobalt(II) in the kinetics of inactivation of aminoacylase by 1,10-phenanthroline and reconstitution of the apoenzyme
AU Wu, Haibin; Tsou, Chenlu
CS Inst. Biophys., Acad. Sin., Beijing, Peop. Rep. China
SO Biochemical Journal (1993), 296(2), 435-41
CODEN: BIJOAK; ISSN: 0306-3275
DT Journal
LA English
CC 7-3 (Enzymes)
AB The kinetics of reconstitution of apoacylase with either Zn(II) or Co(II) and the inactivation of the Co(II)-reconstituted enzyme by 1,10-phenanthroline (OP) has been studied by following the substrate reaction continuously in presence of the metal ion or OP resp. Although the native Zn(II)-contg. and the Co(II)-reconstituted enzymes have closely similar Michaelis consts. and max. velocities, the kinetics for both the inactivation by OP and the reconstitution of the apoenzyme with the metal ions differs considerably. For Co(II), both the inactivation by OP and the reconstitution show simple kinetics, but for Zn(II), the inhibition by OP is a multi-phasic process [Wang, Wu, Wang, Zhou and Tsou (1992) Biochem. J. 281, 285-290] and the kinetics of reconstitution is also much

more complicated. Both the native and the Co(II)-reconstituted enzymes are inhibited by excess of Zn(II), but not by Co(II). The inhibition by Zn(II) in excess and the reconstitution of the apoenzyme with Zn(II) are co-operative processes. The inhibition by Zn and its effect on the fluorescence emission of 1-anilinonaphthalene-8-sulfonic acid bound to the native enzyme indicate multiple Zn(II)-binding sites.

ST **aminoacylase zinc** function binding cobalt probe

IT Kinetics, enzymic
(of **aminoacylase zinc**-contg. and cobalt-substituted derivs, in apoenzyme reconstitution and chelator-mediated inactivation)

IT Conformation and Conformers
(of **aminoacylase**, **zinc** removal and reconstitution effect on)

IT Enzyme functional sites
(metal-binding, of aminoacylase, for zinc, kinetic evaluation of cobalt as probe for)

IT 66-71-7, 1,10-Phenanthroline
RL: BIOL (Biological study)
(aminoacyl native and zinc-substituted deriv. inactivation by, kinetics of)

IT 7440-66-6, Zinc, biological studies
RL: BIOL (Biological study)
(aminoacylase binding sites for and catalytic dependence on, kinetic evaluation of cobalt as probe for)

IT 7440-48-4, Cobalt, biological studies
RL: BIOL (Biological study)
(as probe, for zinc catalytic function and binding sites in aminoacylase, kinetic evaluation of)

IT 9012-37-7, **Aminoacylase**
RL: PRP (Properties)
(**zinc** catalytic function and binding sites in, kinetic evaluation of cobalt as probe for)

L7 ANSWER 3 OF 4 CAPLUS COPYRIGHT 2003 ACS

AN 1992:36826 CAPLUS

DN 116:36826

TI Kinetics of the course of inactivation of aminoacylase by 1,10-phenanthroline

AU Wang, Zhixing; Wu, Haibin; Wang, Xicheng; Zhou, Haimeng; Tsou, Chenlu

CS Natl. Lab. Biomacromol., Acad. Sin., Beijing, 100080, Peop. Rep. China

SO Biochemical Journal (1992), 281(1), 285-90
CODEN: BIJOAK; ISSN: 0306-3275

DT Journal

LA English

CC 7-3 (Enzymes)

AB The kinetic theory of the substrate reaction during modification of enzyme activity previously described (Tsou, C.-L., 1988) has been applied to a study on the kinetics of the course of inactivation of aminoacylase by 1,10-phenanthroline. Upon diln. of the enzyme that had been incubated with 1,10-phenanthroline into the reaction mixt., the activity of the inhibited enzyme gradually increased, indicating dissocn. of a reversible enzyme-1,10-phenanthroline complex. The kinetics of the substrate reaction with different concns. of the substrate chloroacetyl-L-alanine and the inactivator suggest a complexing mechanism for inactivation by, and substrate competition with, 1,10-phenanthroline at the active site.

The inactivation kinetics are single phasic, showing that the initial formation of an enzyme-Zn2+-1,10-phenanthroline complex is a relatively rapid reaction, followed by a slow inactivation step that probably involves a conformational change of the enzyme. The presence of Zn2+ apparently stabilizes an active-site conformation required for enzyme activity.

ST aminoacylase inactivation kinetics phenanthroline; **zinc aminoacylase** inactivation phenanthroline

IT Kinetics, enzymic
(of inactivation, of aminoacylase I of mammal kidney by phenanthroline)

IT 66-71-7, 1,10-Phenanthroline
RL: BIOL (Biological study)
(aminoacylase I of mammal kidney inactivation by, kinetics and mechanism of)

IT 9012-37-7, Aminoacylase I
RL: BIOL (Biological study)
(inactivation of, of mammal kidney by phenanthroline, kinetics and mechanism of)

IT 7440-66-6, Zinc, biological studies
RL: BIOL (Biological study)
(of aminoacylase I, of mammal kidney, in enzymic inactivation by phenanthroline)

L7 ANSWER 4 OF 4 CAPLUS COPYRIGHT 2003 ACS

AN 1988:34088 CAPLUS

DN 108:34088

TI The functional role of zinc in aminoacylase

AU Ou, Yaohua; Zhao, Gang; Zhou, Xin

CS Dep. Biol. Sci. Biotechnol., Tsinghua Univ., Beijing, Peop. Rep. China

SO Shengwu Huaxue Zazhi (1987), 3(5), 411-16
CODEN: SHZAE4; ISSN: 1000-8543

DT Journal

LA Chinese

CC 7-5 (Enzymes)

AB Aminoacylase from pig kidney is a metalloenzyme which contains 2 mol of Zn per mol of protein. The removal of Zn(II) was performed by dialysis of the enzyme (1.2 mg/mL) in 0.17M phosphate buffer, pH 7.3, against 1M 1,10-phenanthroline at 24.degree.. After appropriate time intervals, the Zn contents and the activity were detd. The loss of activity was exactly proportional to the amt. of Zn removed by dialysis. Reactivation of Zn-free inactive aminoacylase was performed by incubation for 20 min at 37.degree. in the presence of 1 .times. 10-4M Zn(II) or Co(II). CD was used to study the effect of Zn on the structural stability of the protein.

The CD spectra were measured with a JASCO J-500 C spectropolarimeter at 18.degree. and 190-240 nm, and the fractions of .alpha.-helix, .beta.-pleated sheet, and random coil in protein were computed with the method of Y. H. Chen et al. (1972). Comparison of the CD spectra of the native aminoacylase and the Zn-free apoenzyme showed that, upon removal of the Zn(II) from the active site, considerable changes in conformation, including an .apprx.7% increase in .alpha.-helix, were induced. Thus, Zn may contribute to the structural stability of the protein. The conformation of the Co(II)-substituted enzyme is similar to the native enzyme, and this may be the reason for restoring of the enzymic activity of metal-free apoenzyme upon the addn. of Co(II).

ST **zinc aminoacylase** conformation

IT Conformation and Conformers

(of **aminoacylase**, **zinc** effect on)
IT 7440-48-4, Cobalt, properties
RL: PRP (Properties)
(conformation of aminoacylase response to)
IT 7440-66-6, Zinc, biological studies
RL: BIOL (Biological study)
(of aminoacylase, function of, in stabilization of enzyme conformation)
IT 9012-37-7, **Aminoacylase**
RL: BIOL (Biological study)
(**zinc** of, function of, in stabilization of enzyme conformation)

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